

NELL-1 promotes cell adhesion and differentiation via Integrin β 1.

Journal:	J Cell Biochem
Publication Year:	2012
Authors:	Jia Shen, Aaron W James, Jonguk Chung, Kyle Lee, James B Zhang, Stephanie Ho, Kevin S Lee, T Mari Kim, Tomoaki Niimi, Shun'ichi Kuroda, Kang Ting, Chia Soo
PubMed link:	22807400
Funding Grants:	Harnessing native fat-residing stem cells for bone regeneration

Public Summary:

NELL-1 (Nel-like molecule-1) is a secreted osteogenic growth factor first identified by its overexpression in human craniosynostosis (CS) patients. NELL-1 protein has been observed to promote bone and cartilage differentiation and to suppress adipogenesis, both in vitro and in vivo. Despite these findings, NELL-1's cell surface receptors have remained unknown. Integrins are cell-surface receptors that mediate inter-cellular adhesion, and adhesion to extracellular matrix structures- important processes for a cell's normal functioning and survival. Previous studies have shown that Integrin β 1-specific collagen-mimetic surface supports osteoblastic differentiation. Recently, NELL-1 has also been verified to promote cell adhesion and osteogenic differentiation but without an understanding of the underlying mechanisms. In this study, we observed for the first time that NELL-1 promotes cell adherence in a dose dependent manner in multiple cell lines; NELL-1 treated cells also exhibited faster adhesion and greater cell spreading. Interestingly, increasing concentrations of NELL-1 led to an increase in cell proliferation, with even the lowest dose producing a significant change. We found that the pre-coating of culture plates with NELL-1 resulted in Integrin β 1 activation, FAK phosphorylation and increased cytoskeletal organization in ST2 murine marrow cells. Importantly, Western blot revealed that NELL-1 directly bound to the extracellular domain of Integrin β 1 and by utilizing siRNA methods, we determined that NELL-1 cell surface binding and enhanced cell attachment were dependent on Integrin β 1 expression. Finally, we observed that pre-coating of culture dishes with NELL-1 resulted in a significant increase in both cell attachment and osteogenic differentiation. To further verify this finding, we employed a 3D model of NELL-1 pre-coating using PLGA (polylactic-co-glycolic acid) scaffolds. Expectedly, cells on pre-coated scaffolds demonstrated increased proliferation and bone formation and mineralization. Our results identify for the first time that NELL-1 directly binds to and activates Integrin β 1, suggesting Integrin β 1 to be the most reasonable cell surface receptor candidate for NELL-1. This binding may be accomplished through the interaction of similar LDVP sequences found on the TSPN domain (Lamin G domain) in both molecules. Still, the specificity of NELL-1 binding to Integrin α remains unknown. A vast number of signaling pathways have been identified to be stimulated by activation of Integrins, including MAPK, and NELL-1 has been previously shown to be able to activate ERK and JNK signaling in multiple cell types. Thus, it is possible that the physical binding of NELL-1 to Integrin β 1 may initiate the activation of MAPK signaling. Lastly, our results reveal greater orthopedic applications and expand on the methods NELL-1 delivery in skeletal tissue engineering. NELL-1 can accelerate bone formation not only by upregulating osteogenic signaling pathways, but also by promoting osteoprogenitor cells' attachment and proliferation via physical interaction with Integrin β 1. Together, our data suggests that NELL-1 can be used to modulate the orthopedic scaffold surface tensional force distribution and therefore to shape tissue morphogenesis.

Scientific Abstract:

NELL-1 (Nel-like molecule-1) is a secreted osteogenic growth factor first identified in human craniosynostosis (CS) patients. NELL-1 protein has been observed to promote bone and cartilage differentiation and to suppress adipogenesis in both in vitro and in vivo models. Despite these findings, the cell surface receptors of NELL-1 have remained unknown. In this study, we observed for the first time that NELL-1 promotes cell adherence in multiple cell lines, including ST2, C3H10T1/2, M2-10B4, ATDC5, and MC3T3 cells. Additionally, we found that NELL-1 binds to extracellular Integrin β 1 and induces cell focal adhesion. By utilizing siRNA methods, we determined that NELL-1 cell surface binding and enhanced cell attachment were dependent on Integrin β 1 expression. Finally, we observed that pre-coating of culture dishes or PLGA (poly lactic-co-glycolic acid) scaffold with NELL-1 resulted in a significant increase in both cell attachment and osteogenic differentiation. Our results identify for the first time a cell surface target of NELL-1, Integrin β 1, and elucidate new functions of NELL-1 in promoting cell adherence and osteogenic differentiation. J. Cell. Biochem. (c) 2012 Wiley Periodicals, Inc.

Source URL: <https://www.cirm.ca.gov/about-cirm/publications/nell-1-promotes-cell-adhesion-and-differentiation-integrinbeta1>